Claims

1. A method for detecting a protein-protein interaction, comprising:

(a) providing a host cell which contains

(i) a first reporter gene operably linked to a DNA sequence comprising a first protein binding site;

(ii) a second reporter gene operably linked to a DNA sequence comprising a second protein binding site;

(iii) a first fusion gene which expresses a first fusion protein, said first fusion protein comprising a first protein covalently bonded to a binding moiety which is capable of specifically binding to said first protein binding site;

(iv) a second fusion gene which expresses a second fusion protein, said second fusion protein comprising a second protein covalently bonded to a binding moiety which is capable of specifically binding to said second protein binding site; and

(v) a third fusion gene which expresses a third fusion protein, said third fusion protein comprising a third protein covalently bonded to a gene activating moiety;

(b) measuring expression output of said first reporter gene as a measure of said interaction between said first and said third proteins;

(c) measuring expression output of said second reporter gene as a measure of said interaction between said second and said third proteins; and

(d) interpreting the expression output results of step (b) and step (c), whereby

(i) increased output of both of said first and said second reporter genes indicates that said third fusion protein interacts with both of said first and said second fusion proteins;

(ii) increased output of said first reporter gene but not said second reporter gene indicates that said third fusion protein interacts with said first fusion protein but not said second fusion protein;

(iii) increased output of said second reporter gene but not said first reporter gene indicates that said third fusion protein interacts with said second fusion protein but not said first fusion protein; and

(iv) no change in output in either of said first or said second reporter genes indicates that said third fusion protein does not interact with either of said first or said second fusion genes.

2. The method of claim 1, further comprising comparing said expression output results of step (b) and step (c) with the expression output result measured in either

(a) a first comparison host cell which contains

(i) a reporter gene operably linked to a DNA sequence comprising a protein binding site;

(ii) a first fusion gene which expresses a first fusion protein, said first fusion protein comprising said first protein covalently bonded to a binding moiety which is capable of specifically binding to said protein binding site; and

(iii) a second fusion gene which expresses a second fusion protein, said second fusion protein comprising said third protein covalently bonded to a gene activating moiety; or

(b) a second comparison host cell which contains

(i) a reporter gene operably linked to a DNA sequence comprising a protein binding site;

(ii) a first fusion gene which expresses a first fusion protein, said

first fusion protein comprising said second protein covalently bonded to a binding moiety which is capable of specifically binding to said protein binding site; and

(iii) a second fusion gene which expresses a second fusion protein, said second fusion protein comprising said third protein covalently bonded to a gene activating moiety; or

(c) both of said first and said second comparison host cells.

3. The method of claim 1, wherein at least one of said first or said second reporter genes may be reduced in expression level.

4. The method of claim 1, wherein one of said first or said second protein binding sites is a tetracycline operator.

5. The method of claim 1, wherein one of said first or said second reporter genes is URA3 or lacZ.

6. The method of claim 1, wherein one of said first or said second reporter genes produces a signal that is received and detected by a second cell.

7. The method of claim 1, wherein said host cell is a yeast cell or a mammalian cell.

8. The method of claim 7, wherein said host cell is a yeast cell.

9. The method of claim 1, wherein said first and said second reporter genes may be expressed simultaneously.

10. The method of claim 1, wherein said first protein and said second proteins are allelic variants.

11. A method for detecting a protein that mediates a change in the state of another protein, comprising:

(a) providing a host cell which contains

(i) a reporter gene operably linked to a DNA sequence comprising a protein binding site;

(ii) a first fusion gene which expresses a first fusion protein, said first fusion protein comprising a first protein covalently bonded to a binding moiety which is capable of specifically binding to said protein binding site; and

(iii) a second fusion gene which expresses a second fusion protein, said second fusion protein comprising a second protein which is capable of interacting with said first protein and which is covalently bonded to a gene activating moiety,

wherein at least one of said first or said second proteins may exhibit a change in state;

(b) allowing said first and said second proteins to interact;

(c) measuring expression of said reporter gene as a measure of said interaction between said first and said second proteins;

(d) introducing into said cell a third gene expressing a third protein;

(e) measuring expression of said reporter gene, a change in said reporter gene expression in the presence of said third protein being an indication that said third protein mediates a change in the state of said first or said second protein leading to an alteration in the ability of said first protein and said second protein to interact.

12. The method of claim 11, wherein said change in state is a conformational change.

13. The method of claim 12, wherein said protein exhibiting a conformational change is a Ras protein.

14. The method of claim 11, wherein said host cell is a yeast cell or a mammalian cell.

15. The method of claim 14, wherein said host cell is a yeast cell.

16. The method of claim 11, wherein the expression of each of said first fusion protein and said third protein occurs in response to an extracellular stimulus.

17. The method of claim 11, wherein said reporter gene produces a signal that is received and detected by a second cell.

18. A method for detecting a protein that mediates a change in the state of another protein, comprising:

(a) providing a first host cell which contains

(i) a reporter gene operably linked to a DNA sequence comprising a protein binding site;

(ii) a first fusion gene which expresses a first fusion protein, said first fusion protein comprising a first protein covalently bonded to a binding moiety which is capable of specifically binding to said first protein binding site;

and

(iii) a second fusion gene which expresses a second fusion protein, said second fusion protein comprising a second protein which is capable of interacting with said first protein and which is covalently bonded to a gene activating moiety,

wherein at least one of said first or said second proteins may exhibit changes in state;

(b) allowing said first and said second proteins to interact;

(c) measuring expression of said reporter gene in said first host cell as a measure of said interaction between said first and said second proteins;

(d) providing a second host cell which contains

(i) said reporter gene operably linked to said DNA sequence comprising said protein binding site;

(ii) said first fusion gene which expresses said first fusion protein; and

(iii) said second fusion gene which expresses said second fusion protein;

(iv) a third gene which expresses a third protein;

(e) allowing said first and said second proteins to interact in the presence of said third protein;

(f) measuring expression of said reporter gene in said second host cell as a measure of said interaction between said first and said second proteins in the presence of said third protein, a change in said reporter gene expression in said second host cell as compared to that measured in said first host cell being an indication that said third protein mediates a change in the state of said first or said second protein resulting in an alteration in the ability of said first protein and said

second protein to interact.

19. The method of claim 18, wherein said change in state is a conformational change.

20. The method of claim 19, wherein said protein exhibiting a conformational change is a Ras protein.

21. The method of claim 18, wherein each of said host cells is a yeast cell or a mammalian cell.

22. The method of claim 21, wherein each of said host cells is a yeast cell.

23. The method of claim 18, wherein the expression of each of said first fusion protein and said third protein occurs in response to an extracellular stimulus.

24. The method of claim 18, wherein said reporter gene produces a signal that is received and detected by a second cell.

25. A cell comprising

(i) a first reporter gene operably linked to a DNA sequence comprising a first protein binding site;

(ii) a second reporter gene operably linked to a DNA sequence comprising a second protein binding site;

(iii) a first fusion gene which expresses a first fusion protein, said first

fusion protein comprising a first protein covalently bonded to a binding moiety which is capable of specifically binding to said first protein binding site;

(iv) a second fusion gene which expresses a second fusion protein, said second fusion protein comprising a second protein covalently bonded to a binding moiety which is capable of specifically binding to said second protein binding site; and

(v) a third fusion gene which expresses a third fusion protein, said third fusion protein comprising a third protein covalently bonded to a gene activating moiety.

26. The cell of claim 25, wherein at least one of said first or said second reporter genes may be reduced in expression level.

27. The cell of claim 25, wherein one of said first or said second protein binding sites is a tetracycline operator.

28. The cell of claim 25, wherein one of said first or said second reporter genes is URA3 or lacZ.

29. The cell of claim 25, wherein one of said first or said second reporter genes produces a signal that is received and detected by a second cell.

30. The cell of claim 25, wherein said host cell is a yeast cell or a mammalian cell.

31. The cell of claim 25, wherein said host cell is a yeast cell.

32. A reporter gene comprising a tetracycline operator operably linked to a gene encoding a detectable product.

33. The reporter gene of claim 32, wherein said gene encoding a detectable product is a URA3 gene or a lacZ gene.

34. A method for detecting whether a candidate protein interacts with a transcriptional activator, comprising:

(a) providing a host cell which contains

(i) a reporter gene which can be reduced in expression level, said reporter gene being operably linked to a DNA sequence comprising a protein binding site;

(iii) a first fusion gene which expresses a first fusion protein, said first fusion protein comprising said transcriptional activator covalently bonded to a binding moiety which is capable of specifically binding to said protein binding site; and

(v) a second fusion gene which expresses a second fusion protein, said second fusion protein comprising said candidate protein covalently bonded to a gene activating moiety;

(b) detecting an increase in expression of said reporter gene as an indication of an interaction between said candidate protein and said transcriptional activator.

35. The method of claim 34, wherein said reporter gene is a URA3 gene.

36. The method of claim 35, wherein said expression level is reduced by 6-azauracil.

37. The method of claim 34, wherein said protein binding site is a tetracycline operator.

38. The method of claim 37, wherein said expression level is reduced by tetracycline or a tetracycline derivative.

39. The method of claim 34, wherein said host cell is a yeast cell or a mammalian cell.

40. The method of claim 39, wherein said host cell is a yeast cell.